

5'~~~~~3' Immunostimulatory oligonucleotide

Immunomers

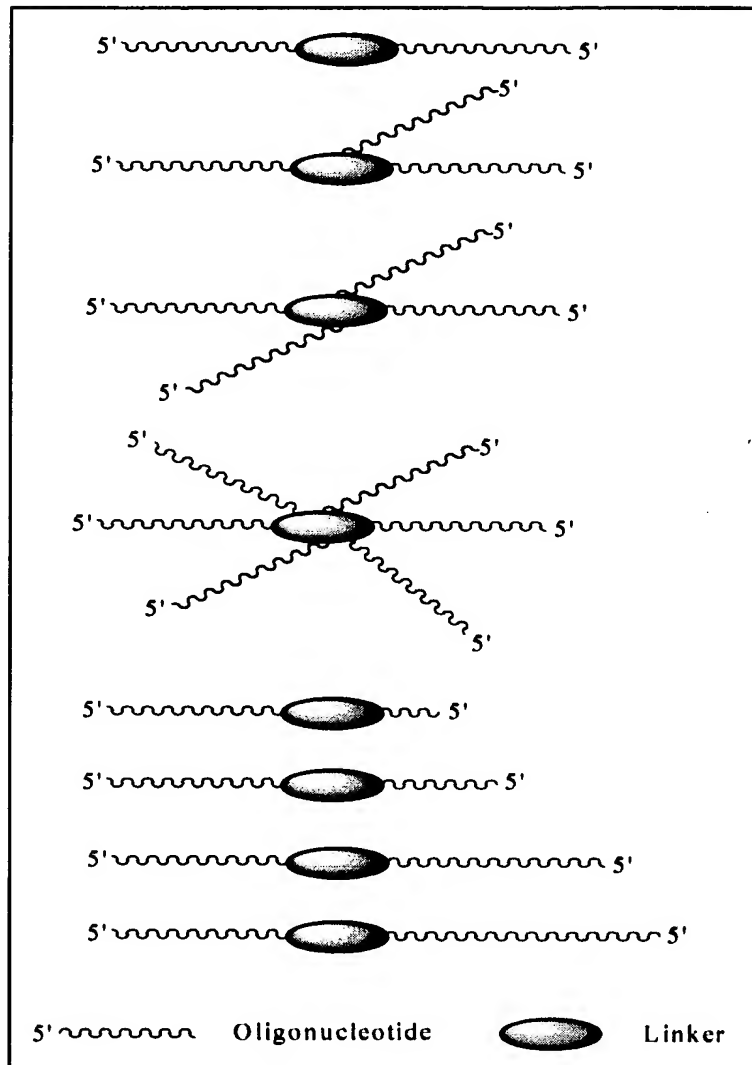
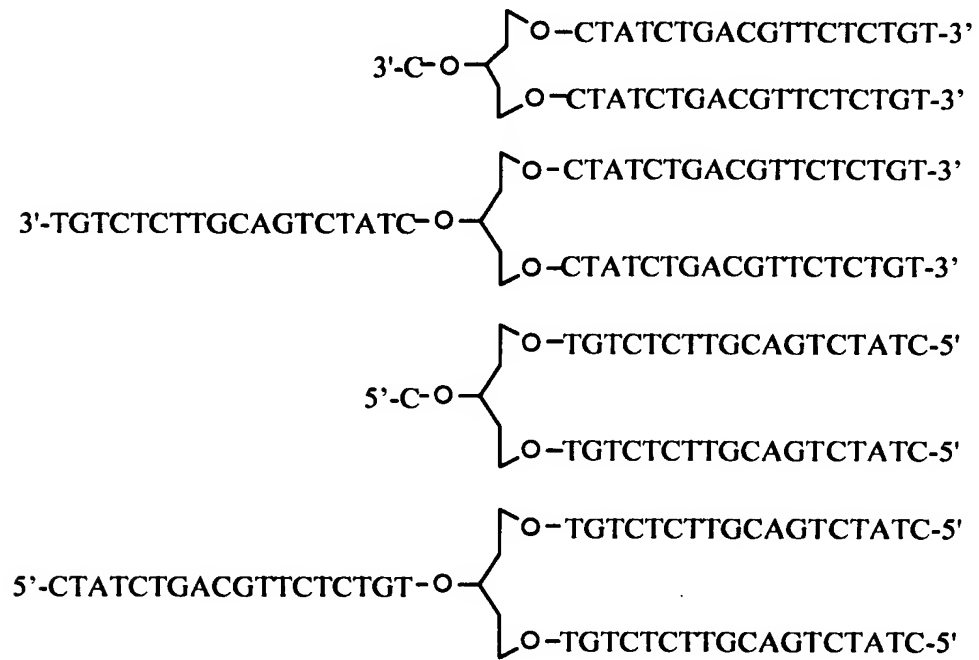


Figure 1

Figure 2



Linkers for linear synthesis

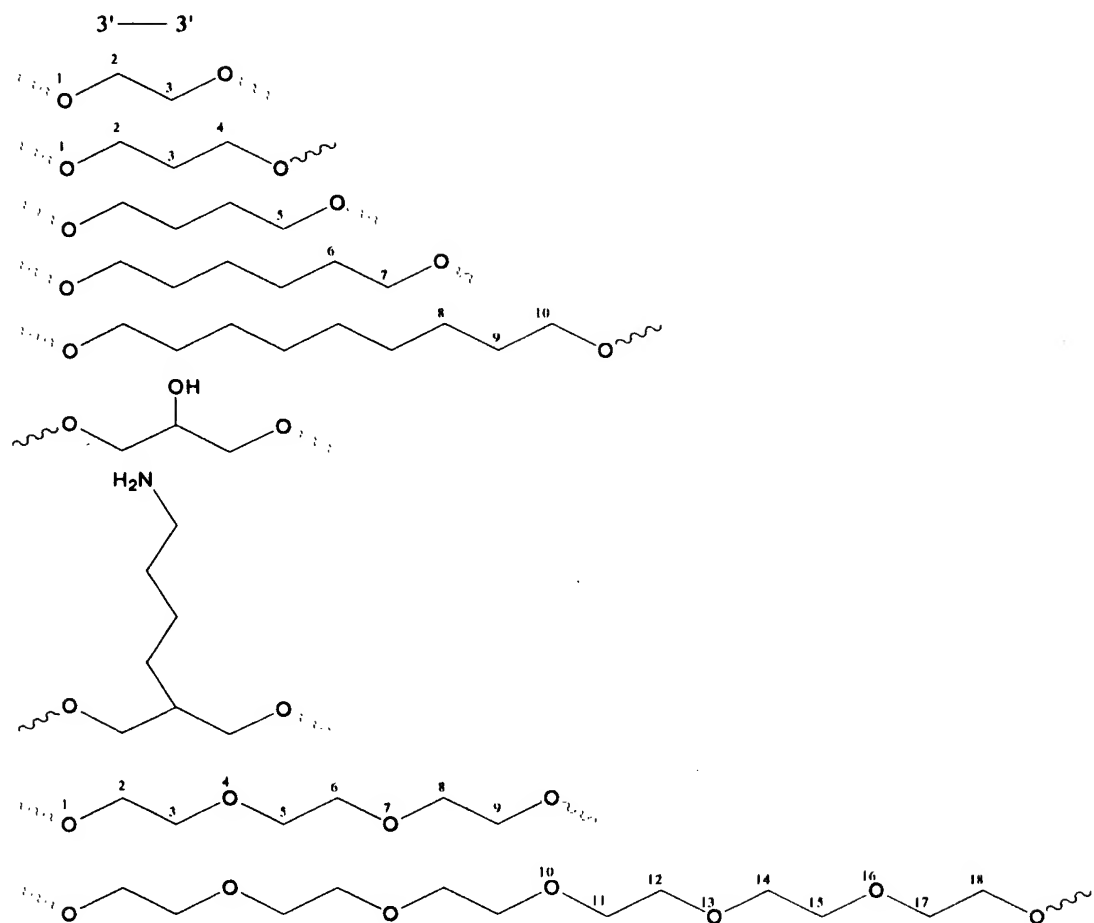
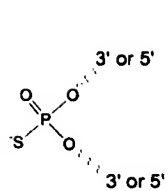
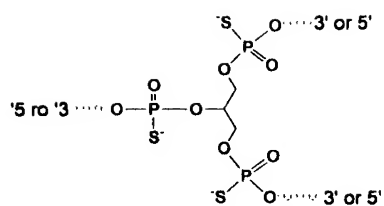


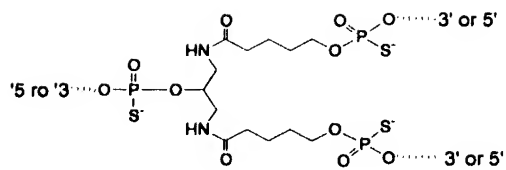
Figure 3



No linker



S, Glycerol brancher;
Short linker



B, Sym. brancher;
Long linker

Figure 4

Linear Synthesis of Immunomers

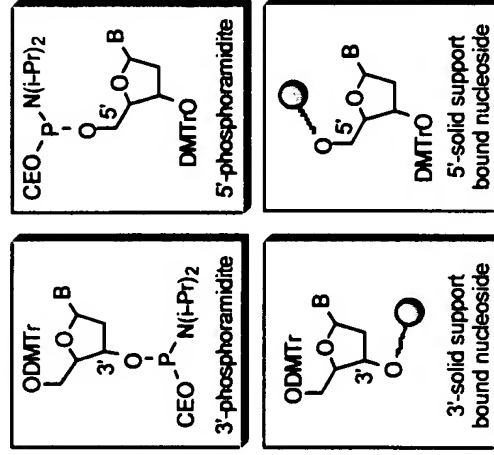
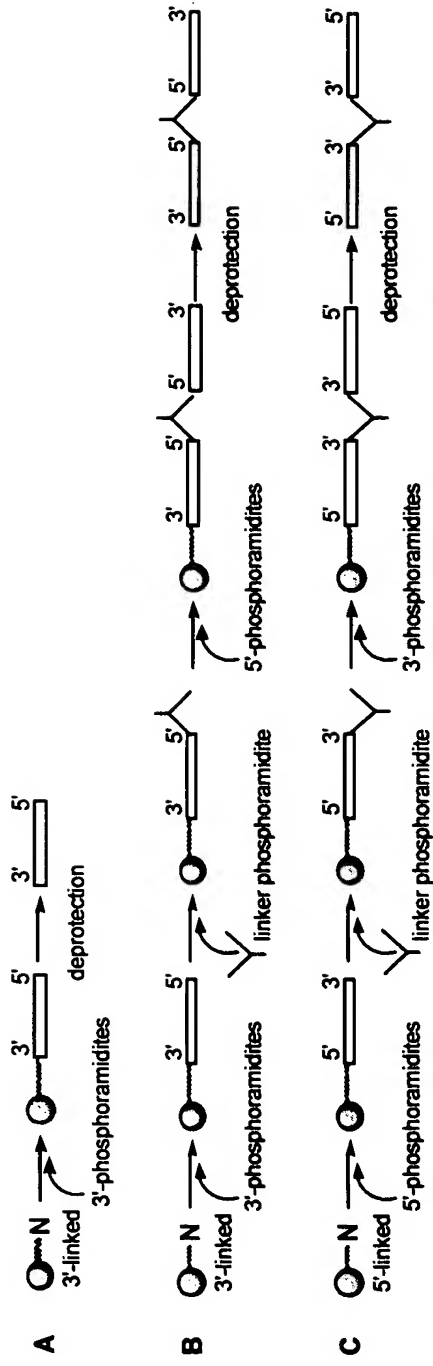


Figure 5

Figure 6

Parallel Synthesis of Immunomers

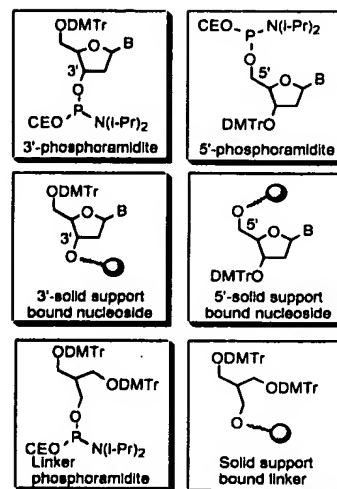
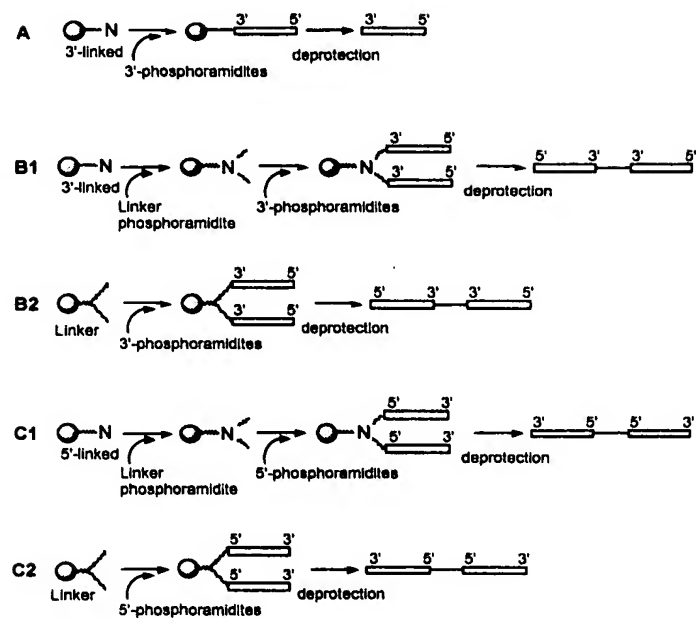


Figure 7

Figure 7A

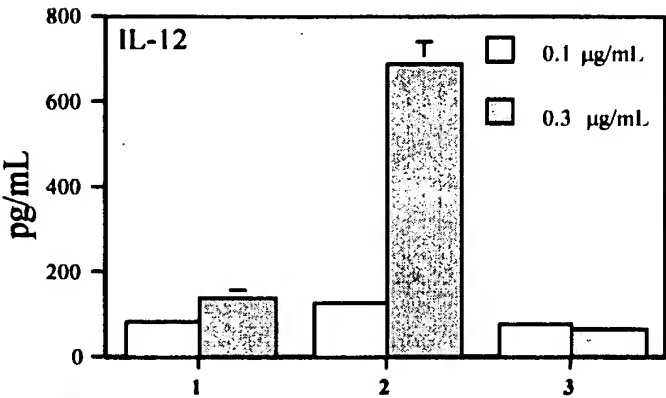


Figure 7B

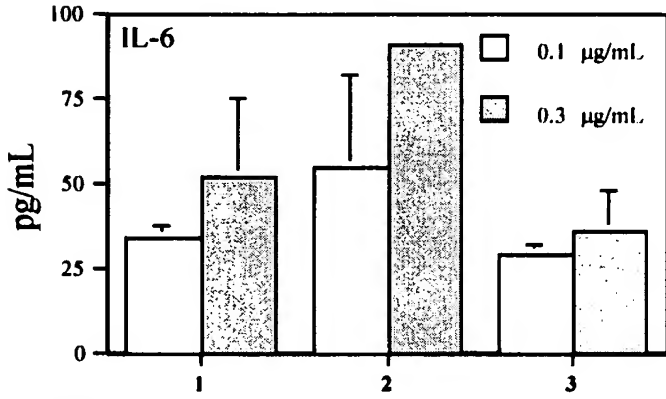


Figure 7C

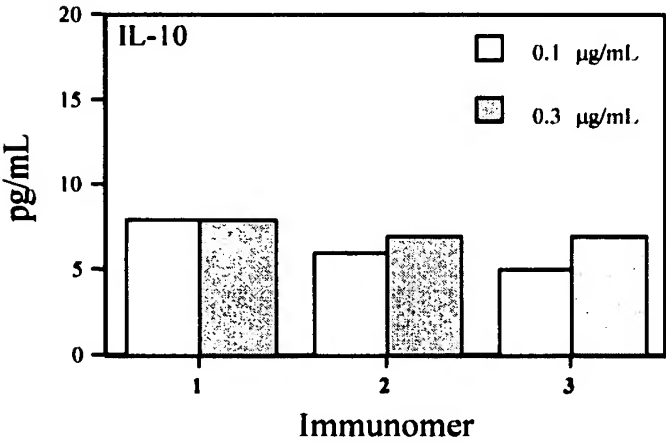


Figure 8.

Figure 8A

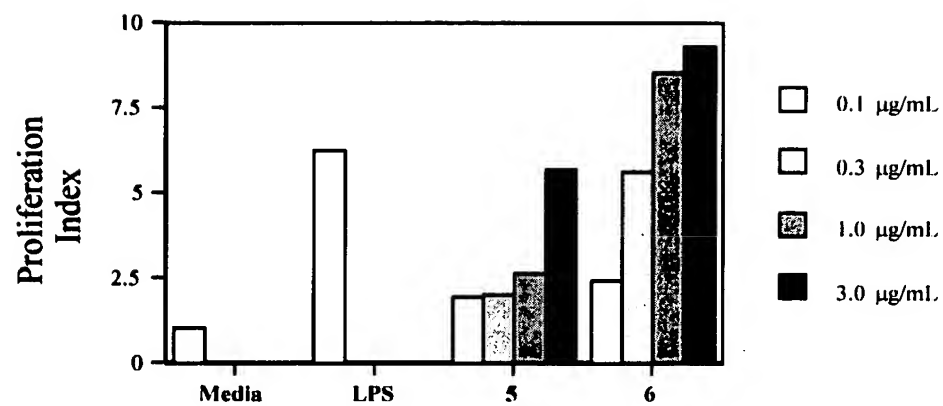


Figure 8B

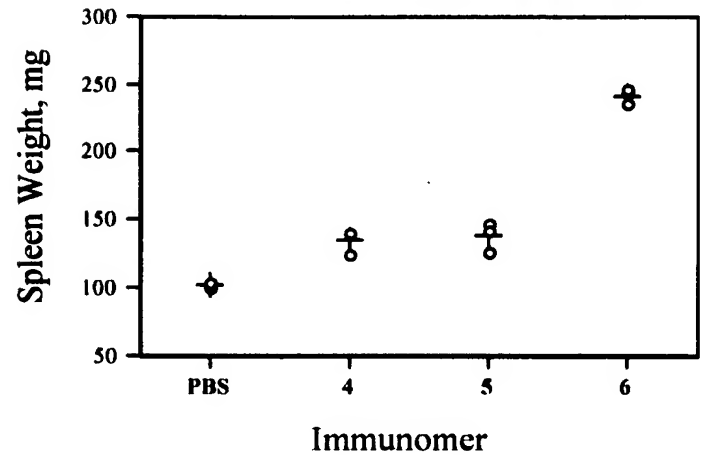


Figure 9.

Figure 9A

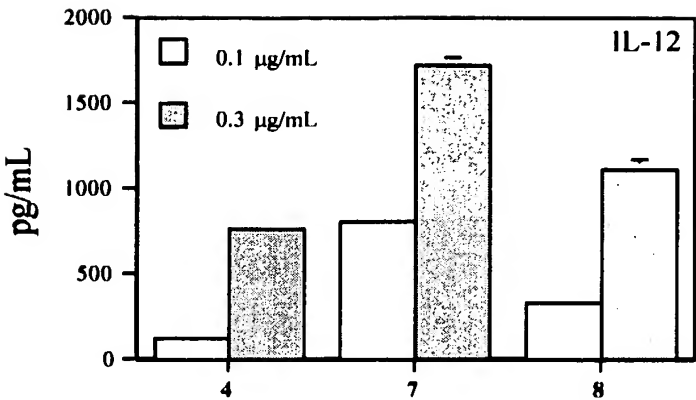


Figure 9B

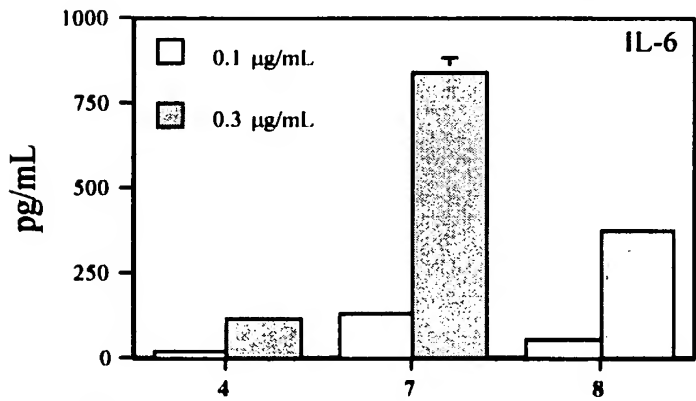
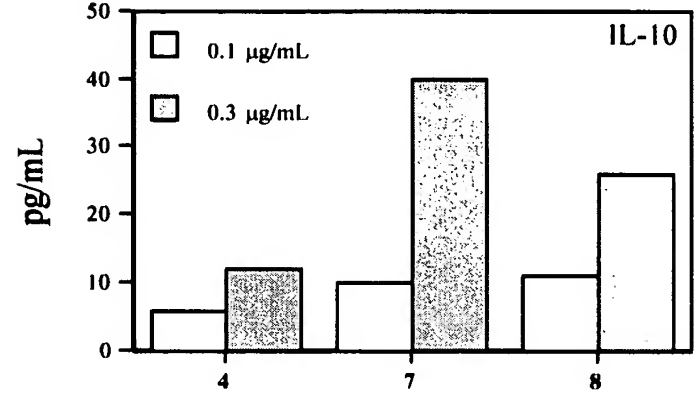


Figure 9C



Immunomer

Figure 10.

Figure 10A

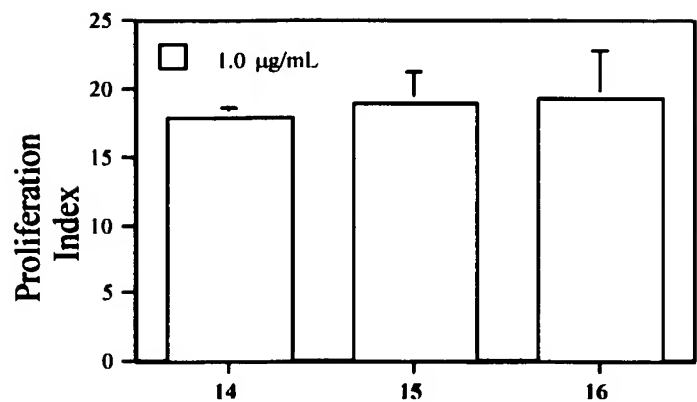


Figure 10B

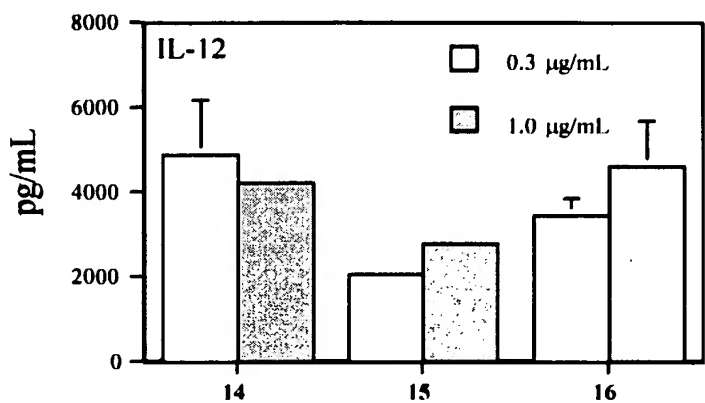


Figure 10C

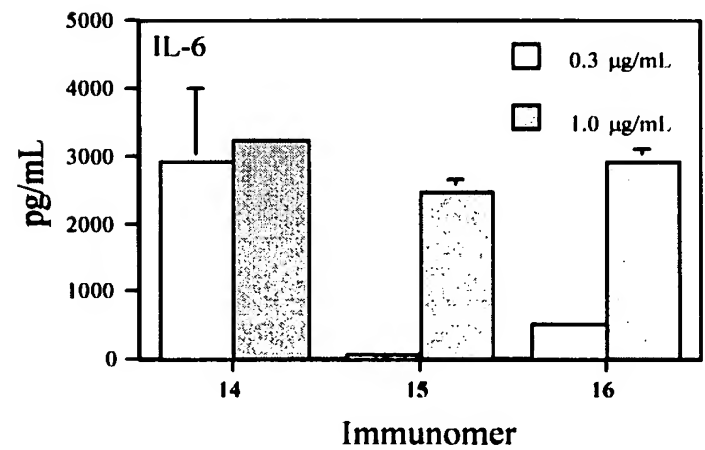


Figure 11.

Figure 11A

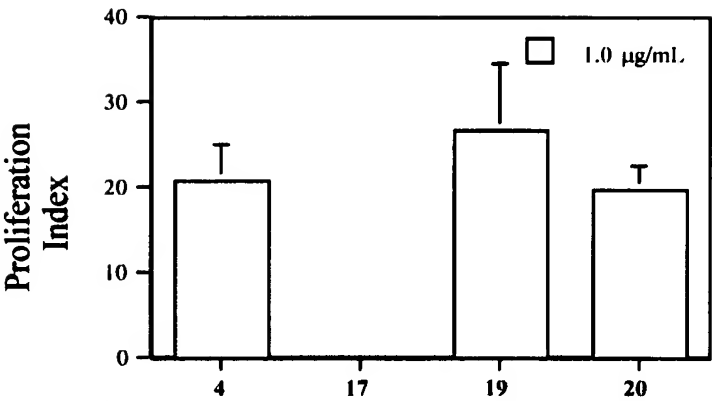


Figure 11B

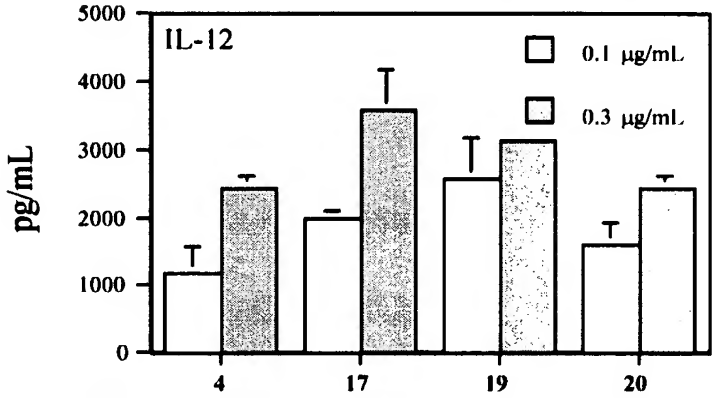


Figure 11C

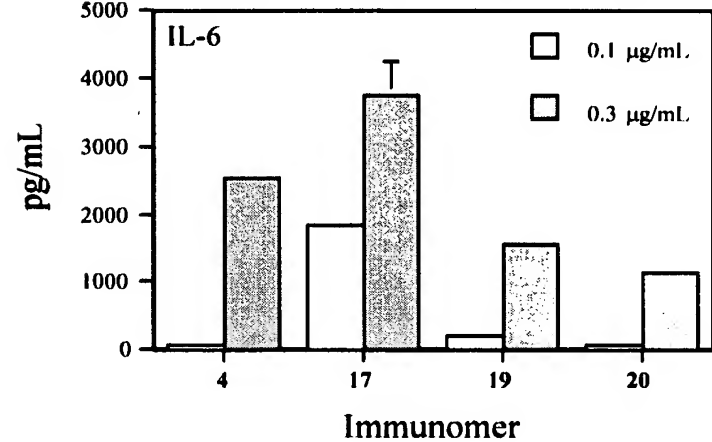
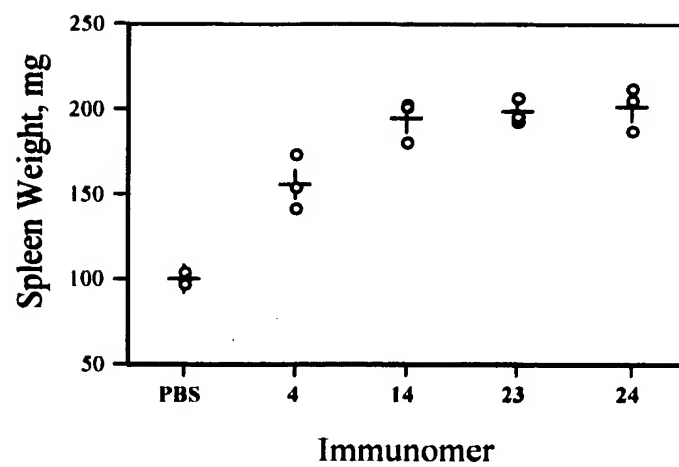


Figure 12.



Possible sites for conjugation

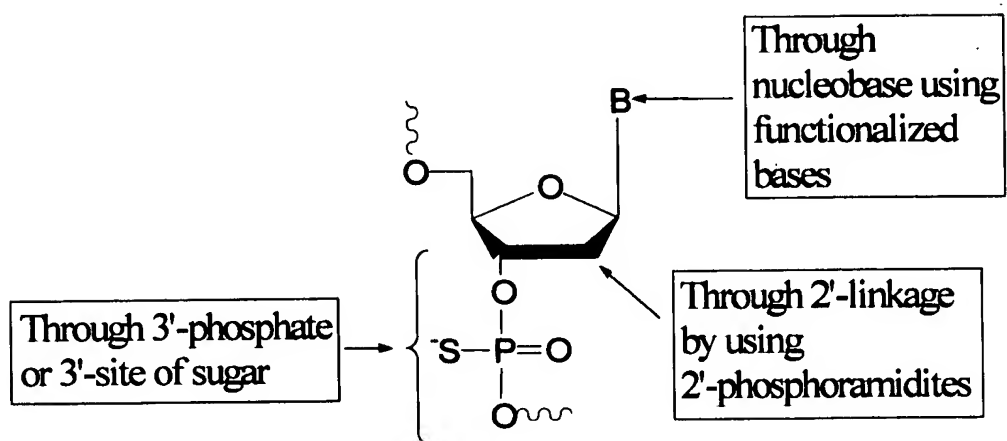


Figure 13

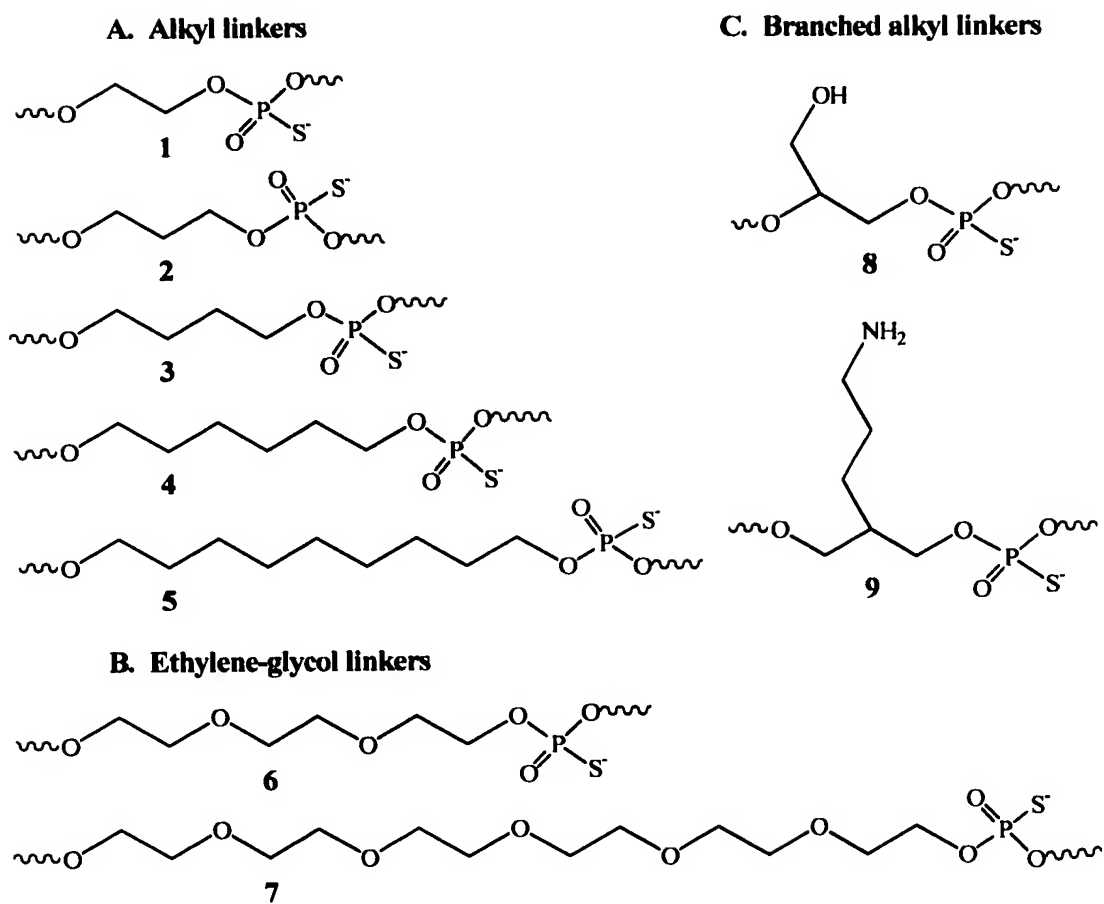


Figure 14

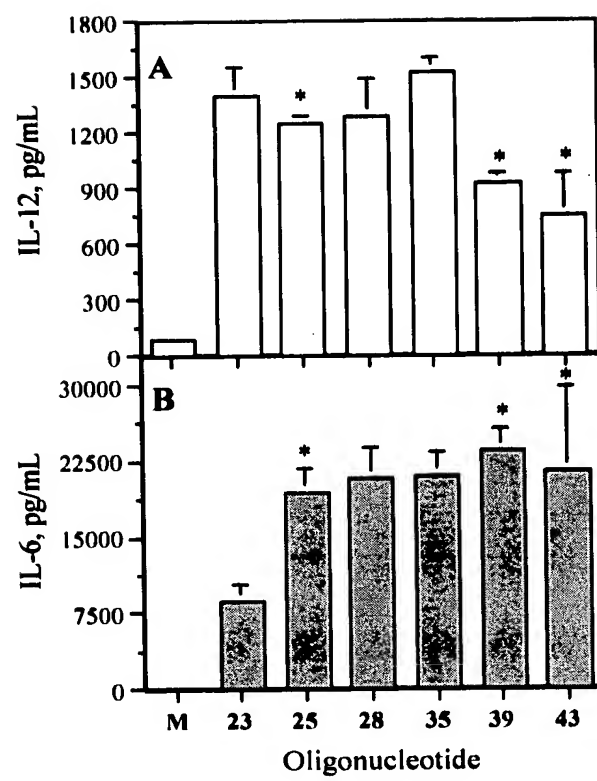


Figure 15

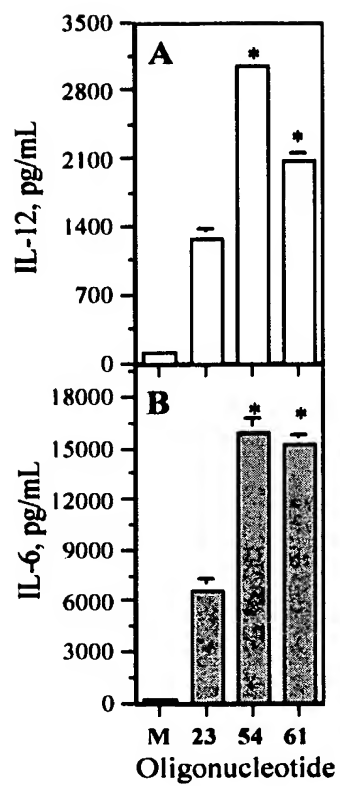


Figure 16

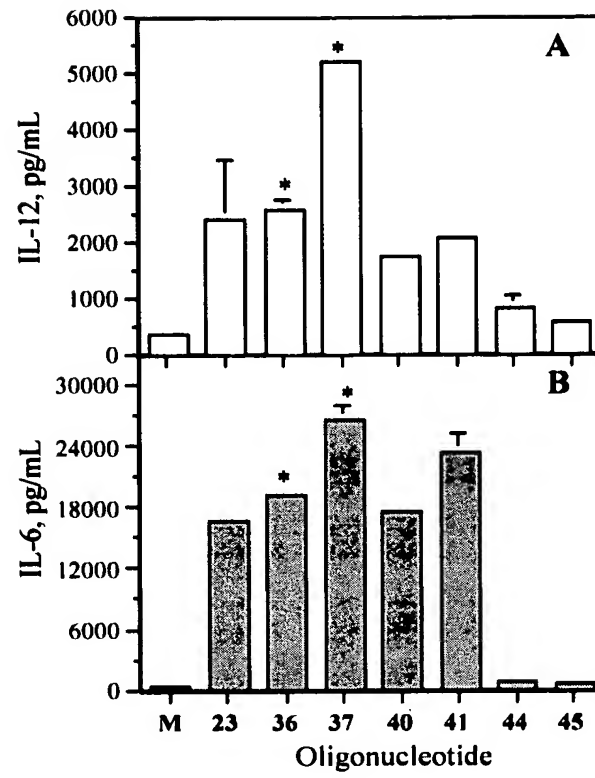


Figure 17

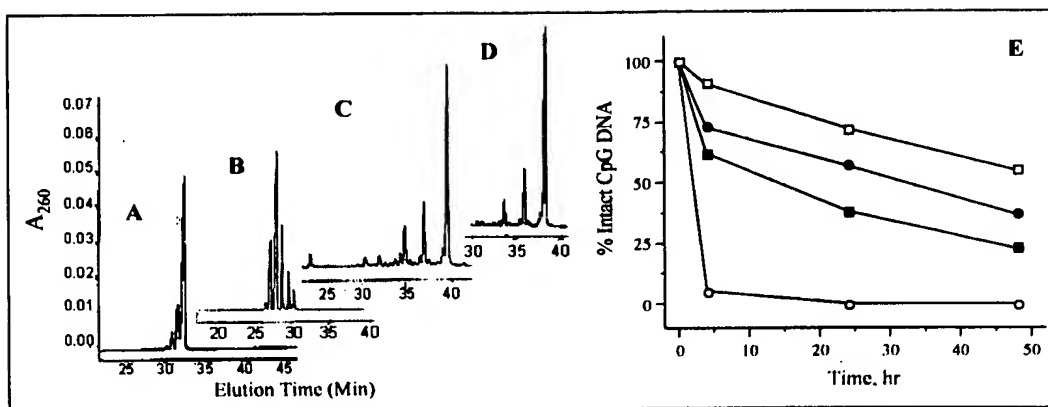


Figure 18

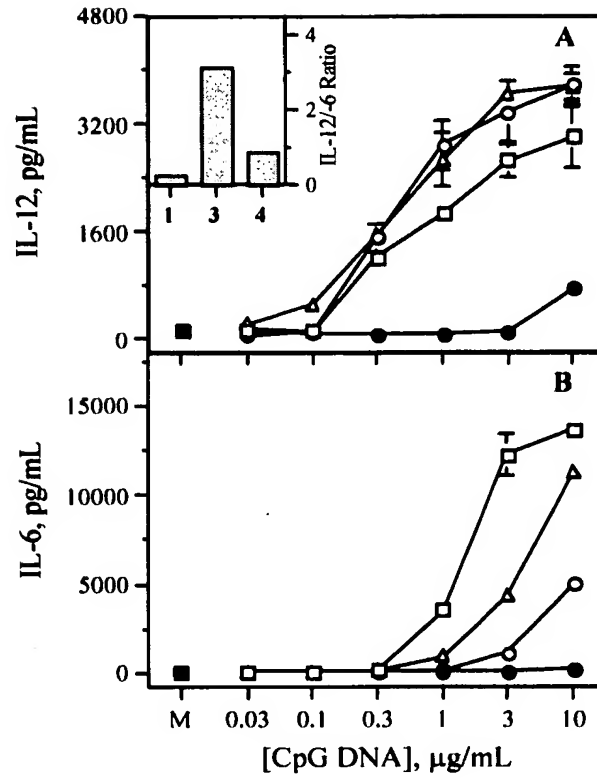


Figure 19

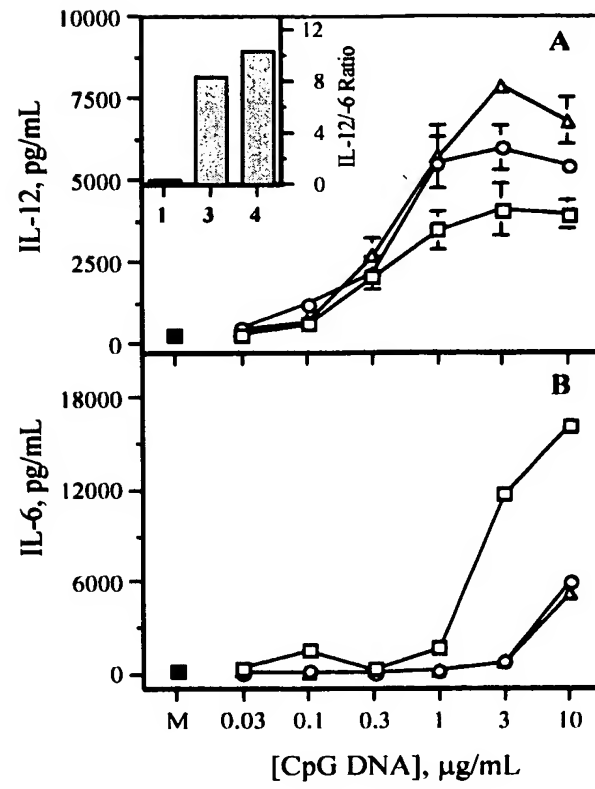


Figure 20

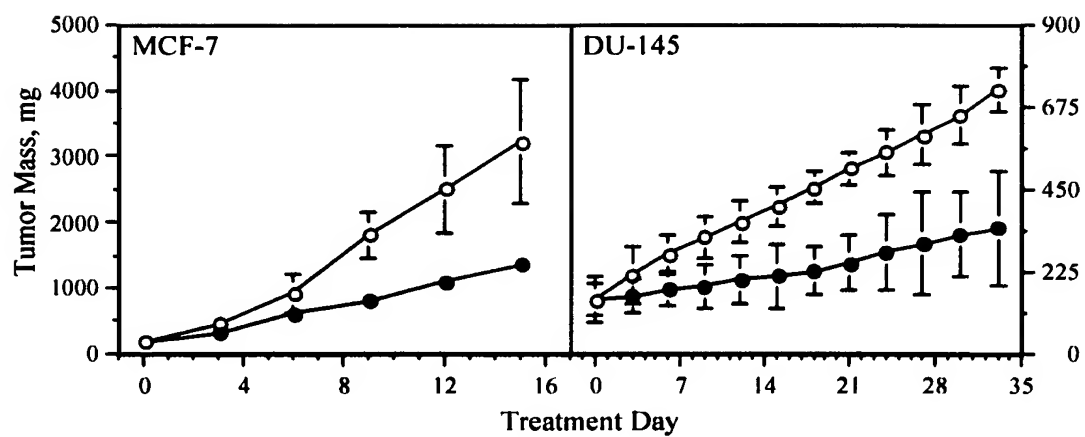
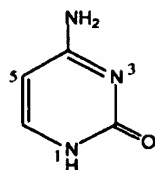
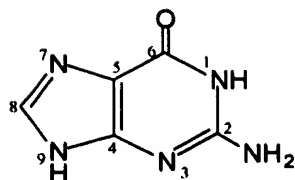


Figure 21

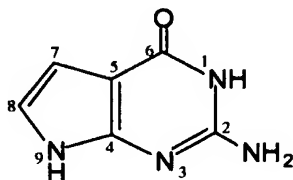
Some pyrimidine and purine structures



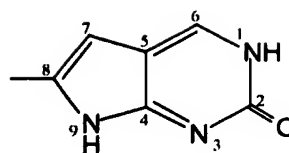
2-Oxy-4-amino pyrimidine or cytosine



2-Amino-6-oxy-purine or Guanine



2-Amino-6-oxy-7-deaza-purine
or 7-Deaza-guanine



2-Oxy-7-deaza-8-methyl purine

Figure 22

- 4 d(5'-CTATCTGACCGTTCTCTGT-3')
 189 d(5'-CTATCTGARGTTCTCTGT-3')
 10 d(5'-CTATCTGACCRTTCTCTGT-3')

 25 d(5'-CTATCTGTCGTTCTCTGT-3')
 190 d(5'-CTATCTGTRGTTCTCTGT-3')

 191 d(5'-TCTGARGTTCT-L-TCTTGRAGTCT-5')
 192 d(5'-TCTGTRGTTCT-L-TCTTGRTGTCT-5')

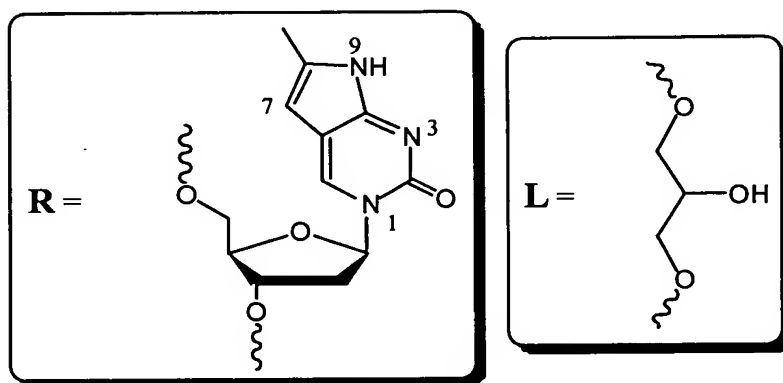


Figure 23

Comparison of Natural Pyrimidine-Purine Immunostimulatory Motif
and Synthetic-Purine-Guanine Immunostimulatory Motif (RpG)

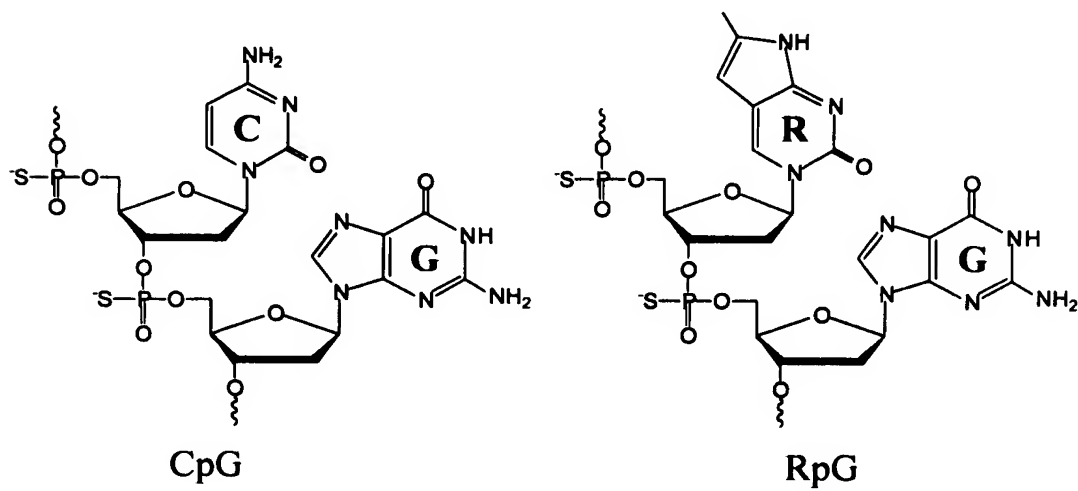


Figure 24

Immunostimulatory activity of parent oligonucleotide 1 containing CpG dinucleotide motif, oligonucleotide 2 containing RpG dinucleotide motif and control oligonucleotide 3 containing GpR dinucleotide motif in mouse spleen cell culture assays. All sequences contain mouse-specific immunostimulatory motif (GACGTT).

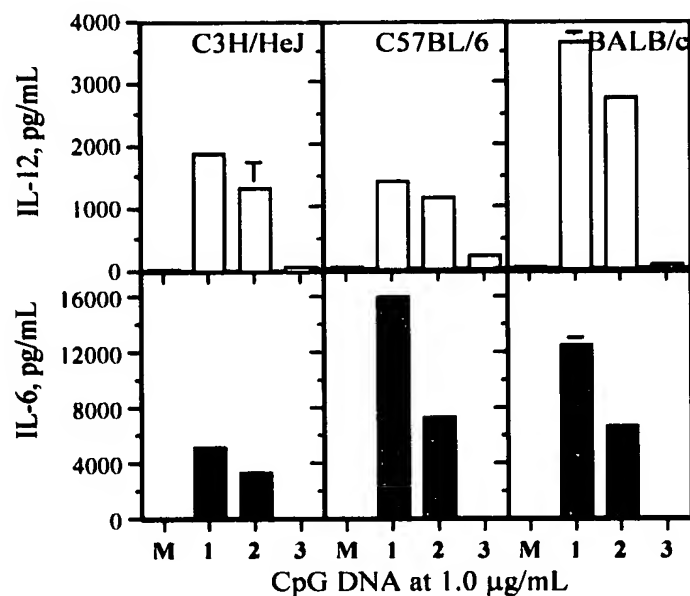


Figure 25

Immunostimulatory activity of parent oligonucleotide **4** containing CpG dinucleotide motif, and oligonucleotide **5** containing RpG dinucleotide motif in mouse spleen cell culture assays. All sequences contain human-specific immunostimulatory motif (GTCGTT).

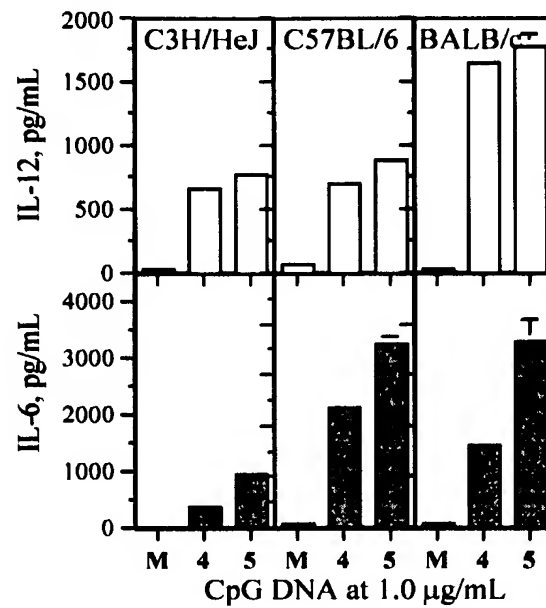


Figure 26

Immunostimulatory activity of parent oligonucleotides **1** and **4** containing CpG dinucleotide motif, and immunomers **6** and **7** containing RpG dinucleotide motif in mouse spleen cell culture assays. Sequences **1** and **6** contain mouse-specific immunostimulatory motif (GACGTT) and sequences **4** and **7** contain human-specific immunostimulatory motif (GTCGTT).

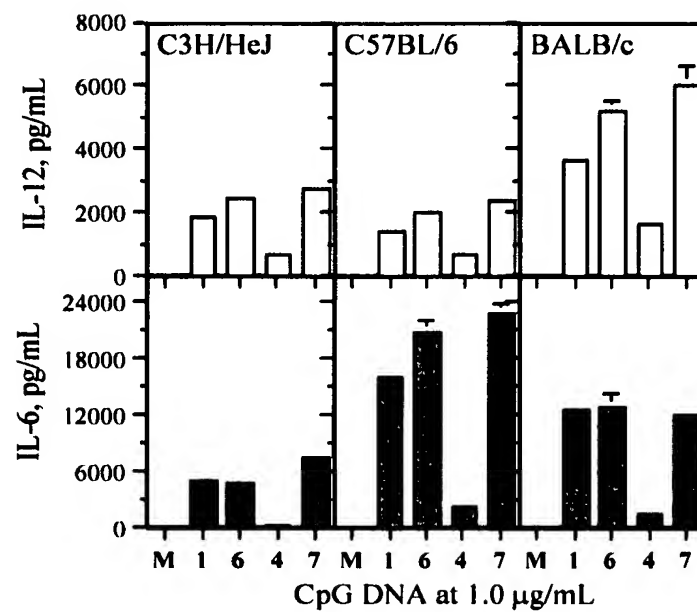


Figure 27

Immunostimulatory activity of parent oligonucleotides **1** and **4** containing CpG dinucleotide motif, and immunomers **6** and **7** containing RpG dinucleotide motif in J774, macrophage-like cell culture assays. Sequences **1** and **6** contain mouse-specific immunostimulatory motif (GACGTT) and sequences **4** and **7** contain human-specific immunostimulatory motif (GTCGTT).

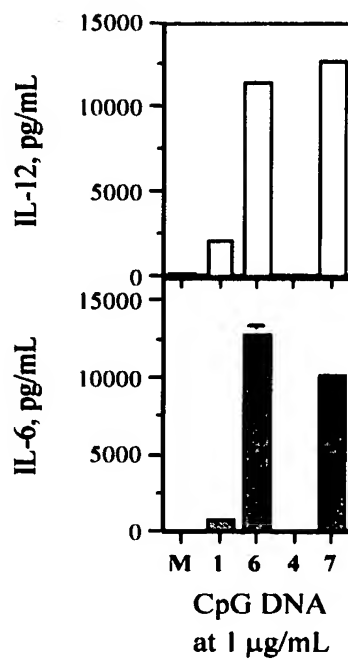


Figure 28

Activation of NF- κ B and degradation of I κ B α in J774 cells as a measure of immunostimulatory activity of parent oligonucleotides 1 - 7.

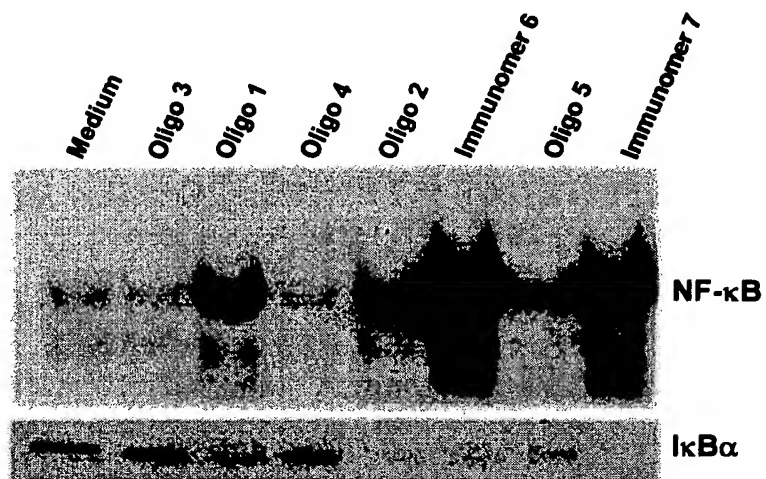


Figure 29

Immunostimulatory activity of immunomer 7 human PBMC cultures (one donor) at 10 $\mu\text{g/mL}$ concentration.

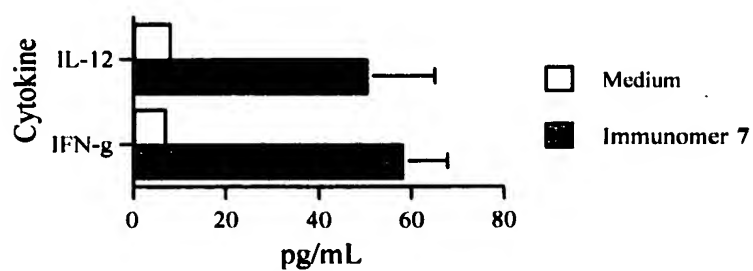


Figure 30